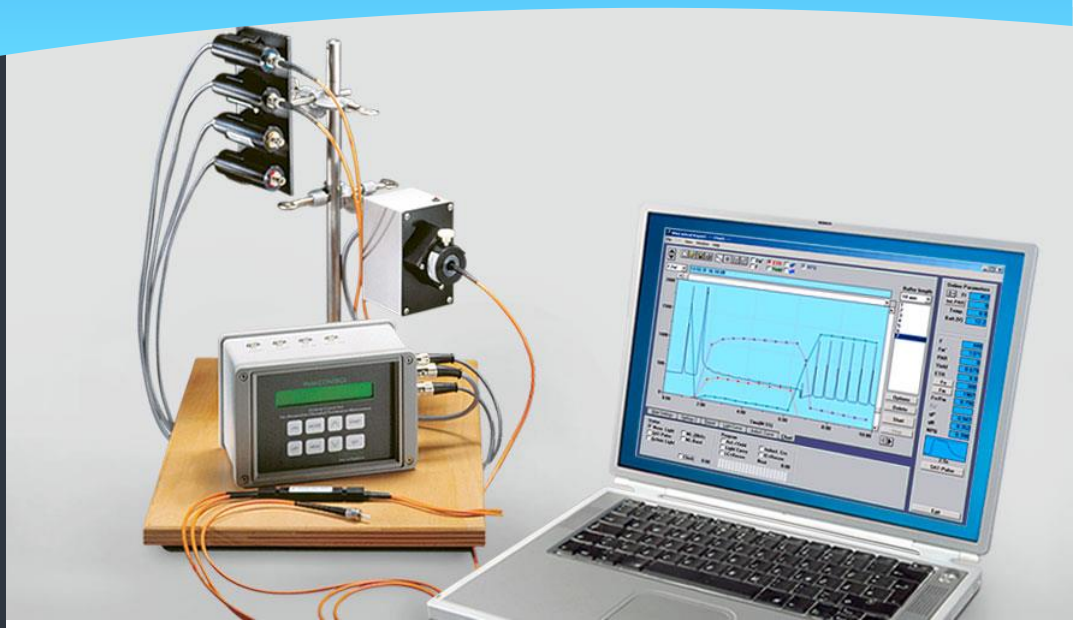
MICROFIBER-PAM: for Probing of Cells and Tissue in Microbial Mats and Leaves



DESCRIPTION

- The MICROFIBER-PAM employs a very thin optical fiber for fluorescence excitation and detection. This narrow fiber permits probing of small spots of heterogeneous photosynthetic surfaces like soil crusts.

- Also, placing the microfiber at different penetration depths permits measurements of photosynthetic gradients within microbial mats and leaves.

MICROFIBER-PAM

General Features

Central part of the MICROFIBER-PAM is a fiber optic coupler consisting of a “beam splitter” to which two pairs of fibers are connected. This “four-port” coupler distributes light incoming from one fiber pair to the other pair where each fiber pair can function as input.

Typically, the fiber optic coupler conducts emission from an LED to the sample, and guides fluorescence from the sample back to a highly sensitive photomultiplier which is shielded against LED light by glass filters.

Both, light emission of the LED and fluorescence measurement by the photomultiplier are coordinated by the PAM-CONTROL unit.

The PAM-CONTROL unit allows stand-alone operation of the MICROFIBER-PAM but functions also as a physical interface for computer-controlled operation of the MICROFIBER-PAM.

Normally, the MICROFIBER-PAM uses blue LED light for fluorescence excitation but alternative LEDs emitting in the green and red spectral range are available.

Peak emission of the blue LED is 470 nm. The LED emits pulses of several µs duration to elicit the pulse-modulated fluorescence measured by the MICROFIBER-PAM , but also longer-lasting pulses which produce integrated light intensities which can saturate photosynthesis.

MICROFIBER-PAM

ACCESSORIES

Working Fiber MF-F

For fluorescence measurements, one of the fibers opposite to the LED/photomultiplier fibers is placed in close contact to the sample, providing output of excitation light and input of fluorescence. Also, this fiber can be connected to a “working fiber” (MF-F) which is a single fiber with one free end of 100 µm diameter.

Green (MF-L520) and Red (MF-L630 and MF-L650) Measuring Light LEDs

Fluorescence excitation by blue light is inefficient in many cyanobacteria. Therefore, we offer two types of red LEDs for studies of cyanobacteria using the MICROFIBER-PAM.

Functional Schematic

- The blue light is passed through a short-pass filter transmitting only at wavelengths below 600 nm (MF-L470). The LED is coupled via ST connection to one fiber of the four-port fiber optic coupler, MF-2-2-100. To the parallel fiber of the MF-2-2-100 coupler, the photomultiplier (PM-MF) is attached using an adapter.
- The adapter for the photomultiplier is part of the MICROFIBER-PAM adapter set, MF-A. The MF-A adapter set also includes a holder for the blue LED and three additional LEDs.
- The photomultiplier is shielded against the blue LED light by long-pass filters transmitting light only at wavelength greater than 640 nm. Without these filters, and face to face orientation of the tips of the two output fibers, the photomultiplier predominantly detects the blue measuring light transmitted between output fibers.
- Obviously, a light absorbing sample placed between the fiber tips reduces the blue light intensity reaching the photomultiplier. Hence, when the MICROFIBER-PAM is configured to detect measuring light, the degree of light attenuation by a sample can by assessed from the photomultiplier signal**.**
- Both, LED and photomultiplier are connected to PAM-CONTROL unit. The PAM-CONTROL unit is delivered with WinControl V.2-Software for operation by Windows computers. a charger MINI-PAM/L, a cable to connect a chart recorder and a transport box.

Application

- The investigation of gradients of photosystem II characteristics within the leaf. For such investigations, the pointed tip of a working fiber (MF-F) is advanced into the leaf tissue by a micromanipulator, and saturation pulse analysis is carried out at defined depths of penetration**.**
- Analyzing how strong light of different colors damages photosystem II at various distances from the leaf surface. By measuring the maximum photochemical quantum yield of photosystem II (FV/FM), the authors demonstrated that amplitude as well as leaf gradients of photoinhibition depend on the color of photoinhibitory light

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